



A dynamic model to calculate cadmium concentrations in bovine tissues from basic soil characteristics

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ABSTRACT

A chain model was developed to calculate the flow of cadmium from soil, drinking water and feed towards bovine tissues. The data used for model development were tissue Cd concentrations of 57 bovines and Cd concentrations in soil, feed and drinking water, sampled at the farms where the bovines were reared. Validation of the model occurred with a second set of measured tissue Cd concentrations of 93 bovines of which age and farm location were known. The exposure part of the chain model consists of two parts: (1) a soil–plant transfer model, deriving cadmium concentrations in feed from basic soil characteristics (pH and organic matter content) and soil Cd concentrations, and (2) bovine intake calculations, based on typical feed and water consumption patterns for cattle and Cd concentrations in feed and drinking water. The output of the exposure model is an animal-specific average daily Cd intake, which is then taken forward to a kinetic uptake model in which time-dependent Cd concentrations in bovine tissues are calculated. The chain model was able to account for 65%, 42% and 32% of the variation in observed kidney, liver and meat Cd concentrations in the validation study.

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1. Introduction

A recent survey in Belgium showed that almost 50% of kidneys and 4% of livers from cattle reared in rural (uncontaminated) areas and 75% of kidneys and 25% of livers from cattle reared in contaminated areas, exceeded the European maximum levels for cadmium (Cd) of 1 and 0.5 mg kg^{−1} fresh weight respectively (Waegeneers et al., 2009a). The examined bovines were 0.5 to 11.5 years old. As the Cd concentration in kidneys increases with age (e.g. Andersen and Hovgård Hansen, 1982; Lindén et al., 1999) the age of the animals was assumed to play an important role in the high number of samples exceeding the European maximum level. Further calculations showed, however, that already in two year-old bovines more than 5% of kidneys would exceed the maximum level in rural areas (Waegeneers et al., 2009b). Kidneys from known contaminated areas are generally banned from the human food chain in different countries. However, there is no guarantee that tissues from other areas do comply with the maximum levels. Monitoring is the most comprehensive method to determine whether tissues comply with maximum levels. It is, however, too expensive and time consuming to analyze all kidney and liver samples before their further commercial use. Modeling the transfer of Cd through the soil–feed–animal tissue food chain is an interesting tool that could help identifying farms or regions that are

susceptible for exceedance of maximum limits due to local soil characteristics. Given the fact that Cd levels in animal tissues increase with age (Andersen and Hovgård Hansen, 1982; Waegeneers et al., 2009b), the time aspect has to be incorporated in this modeling.

Dynamic models take into account the age-dependency of tissue Cd concentrations and are very useful in predicting the increase in Cd levels in animal tissues with time, but they are scarcely available for grazing animals and are often not based on or validated with paired soil–feed–animal tissue concentrations. Chain models that include this time aspect were developed by Loganathan et al. (1999) for sheep and by Franz et al. (2008) for cattle. Both these authors used linear relations to calculate the transfer of Cd from feed, soil and water to animal tissues, assuming no excretion of Cd from these tissues. It is, however, most likely that Cd is excreted from liver (Chan and Cherian, 1993) and kidneys (Smith et al., 1991; Beresford et al., 1999). Beresford et al. (1999) developed a feed–tissue transfer model for sheep which takes into account Cd excretion, because there was a need to be able to estimate how rapidly an animal will decontaminate once the source of contamination is removed from the diet. The model of Beresford was adapted to dairy cattle by Crout et al. (2004).

The objective of this study was to develop a dynamic soil–feed–animal tissue chain model for Cd, incorporating soil–plant transfer models, bovine intake calculations and a kinetic uptake model. The chain model differs from that of Franz et al. (2008) in two points: it incorporates a kinetic model for the metabolism of Cd, and it is based on measured tissue Cd concentrations from animals with a known history and with corresponding measured Cd concentrations in soil, feed and drinking water.

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2. Materials and methods

2.1. Sample collection and analysis

Kidneys, livers and meat tissue of 150 bovine animals were sampled in 2005 by the Belgian Federal Agency for the Safety of the Food Chain (FASFC) and analyzed for cadmium, lead, arsenic, zinc and copper (Waegeneers et al., 2009a). The samples originated from both uncontaminated areas (so-called reference areas) and contaminated areas. A more detailed description of the areas and a geographical location are given in Waegeneers et al. (2009a). Together with the samples of each bovine, a copy of the bovine passport was received, containing information on the date of birth, sex and type of the animal (dairy or beef) and on the farms where the animal was reared. Farms where the animals had resided for more than 18 months, were contacted and the farmers were asked to further participate voluntarily. In total, 53 farmers responded positively, covering 57 animals. These 57 cases were studied in depth: the corresponding farms were visited once in spring, once in summer and once in autumn in the period 2006–2007. A questionnaire was filled out with the farmers, rendering information about the feed supplied to the animals, the type of drinking water the animals had access to, the location of pastures and agricultural fields from where the locally cultivated feed originated, and the time spent indoors (stable) and outdoors (pasture) by the animals. Samples of fresh pasture grass, silage grass, hay, maize, soil and drinking water were collected at these farms.

Fresh pasture grass was sampled in spring, summer and autumn by clipping at least 20 subsamples of grass at random throughout each of the pastures on which the animals had grazed. Silage grass, hay and maize were collected by grabbing randomly at least at five places in the respective silos. The surface layer of 127 pasture soils (0–10 cm) was sampled with a grass plot sampler, 135 arable soils were sampled with a gauge auger (0–30 cm). The soils were selected based on the information given in the questionnaires (i.e. soils on which grass and maize from the silages had grown + from pastures on which the bovines had grazed). To give representative samples of each field, 10–20 soil cores were collected and bulked in the field. Well water and surface water were also sampled if they served as a source of drinking water in the stable or in the pastures. Tap water from the water distribution net was not sampled. The water was collected in 250-ml plastic jars and acidified immediately with nitric acid.

Post harvest, all grass samples (unwashed) were cut in small pieces (maximum 1 cm length), carefully mixed by hand to homogenize the sample, and part of it (200–500 g) was dried at 70 °C until constant weight. Silage samples were also carefully mixed by hand, and a subsample (200–600 g) was dried at 70 °C. After drying and grinding with a hammer mill, all vegetal samples were wet digested (4 ml concentrated HNO₃ + 4 ml demineralised water) in a microwave (MarsXpress, CEM Corporation, Matthews, NC, USA). Cadmium concentrations in the extracts were measured by ICP-MS (inductively coupled plasma-mass spectrometry; VG PQ-ExCell Thermofisher Scientific, Winsford, UK). Each batch of 10 samples additionally included one procedure blank, one reference material

(IAEA-407, International Atomic Energy Agency, Vienna, Austria) and one laboratory control material. The limit of quantification (LOQ) was calculated as 10 times the standard deviation of ten procedure blanks, multiplied with the dilution factor and equalled 2 µg kg⁻¹.

All soil samples were passed through a 2-mm sieve and dried at room temperature for several weeks. Cadmium concentrations in soil were determined by ICP-OES (Liberty 2000, Varian Inc., Palo Alto, USA) after aqua regia extraction. Each batch of 10 samples additionally included one procedure blank, two certified reference materials (CRM 7002—Light sandy soil and CRM 7004—Loam soil, Analytika Co. Ltd., Prague, Czech Republic) and one laboratory control material. The LOQ value expressed on a dry weight basis was 0.10 mg kg⁻¹. The organic matter content was determined by the Walkley & Black method (Jackson, 1958). Soil pH was measured in distilled water (1:2.5 soil/solution ratio) after stirring for 15 min.

Statistical analyses of soil and plant data were performed with UNISTAT Statistical Package, Version 5.6 (UNISTAT Ltd, London, UK). The normal distribution of data was verified by the Kolmogorov–Smirnov test with Lilliefors correction. Data that were log-normally distributed, were log₁₀-transformed before further analysis. Data were analyzed by one-way analysis of variance (ANOVA) followed by the Tukey–HSD multiple range test ($\alpha = 0.05$). Basic statistics were calculated on untransformed data.

2.2. Modeling concepts

2.2.1. Soil–plant transfer

Soil–plant regression models were derived to estimate Cd concentrations in locally produced grass and maize (silage) from soil characteristics (Cd concentration in soil, pH and organic matter content). The regression model used is presented in Eq. (1):

$$\log_{10}(\text{Cd} - \text{plant}) = a + b * \log_{10}(\text{Cd} - \text{soil}) + c * \text{pH} + d * \log_{10}(\text{organic matter}) \quad (1)$$

with Cd-plant and Cd-soil in mg kg⁻¹ on a dry weight basis and organic matter expressed in percentage. A logarithmic transformation of data was necessary to obtain a normal distribution. The regression parameters were derived by stepwise multiple linear regression (UNISTAT 5.6; $\alpha = 0.05$ to enter a variable and $\alpha = 0.10$ to remove a variable) with the plant and soil data from the 57 cases that are studied in-depth. The significant regression parameters can be found in Table 1.

2.2.2. Bovine intake of trace elements

The exposure model concept is based on the cadmium exposure model by Römken et al. (2007) and is presented in Fig. 1. The input parameters of the model are trace element (TE) concentrations in grass, maize, soil, drinking water and in animal feed that is not locally produced (mainly concentrates). Whenever TE concentrations in grass and maize are not available, they can be estimated with Eq. (1) and the parameters presented in Table 1.

The lifetime exposure of bovines to Cd is calculated as the sum of Cd exposures during three different life stages (calves and weaners up

Table 1

Regression parameters for significant Cd soil–plant regression equations of the type $\log_{10}(\text{Cd-plant}) = a + b * \log_{10}(\text{Cd-soil}) + c * \text{pH} + d * \log_{10}(\text{OM})$.

		n	Intercept a	Cd-soil b	pH c	OM ^a d	R _{adj} ²
Grass	Spring pasture	117	0.63	0.79 ± 0.06	−0.16 ± 0.04	−0.71 ± 0.13	0.59
	Summer pasture	115	0.25	0.71 ± 0.06	−0.15 ± 0.05	–	0.54
	Autumn pasture	108	−0.38	0.77 ± 0.06	–	−0.34 ± 0.14	0.63
	Hay/silage	46	−0.62	0.66 ± 0.16	–	–	0.29
Maize	Silage	63	1.83	0.71 ± 0.16	−0.32 ± 0.08	−1.11 ± 0.27	0.37

^a OM is the organic matter content.

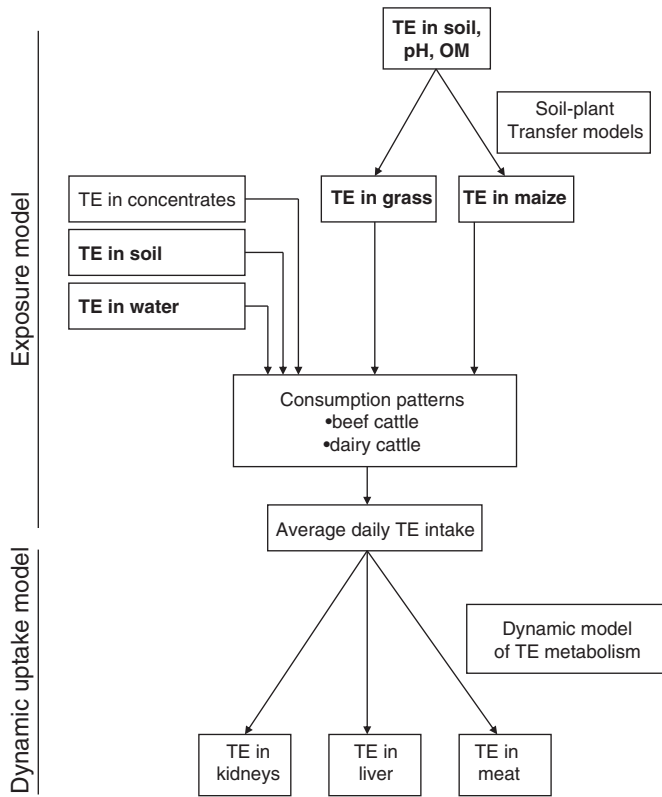


Fig. 1. Chain model to calculate trace elements (TE) in kidneys, liver and meat of bovines, based on the total intake of these elements by the cattle.

to the age of 1 year; yearlings up to the age of 2 years; bovines older than 2 years) as proposed by Römken et al. (2007) and Franz et al. (2008):

$$\text{Lifetime exposure } (\mu\text{g}) = \Delta t_{\text{calf}} \cdot \sum (I_i \cdot C_i) + \Delta t_{\text{yearling}} \cdot \sum (I_i \cdot C_i) + \Delta t_{\text{adult}} \cdot \sum (I_i \cdot C_i) \quad (2)$$

with i standing for the different ingested matrices (feed, water, and soil), I_i the ingested amount of matrix i (in kg dry matter day^{-1}), C_i the Cd concentration in matrix i (in $\mu\text{g kg}^{-1}$ dry matter), and Δt the time (in days) spent as a calf/weaner, yearling and “adult” bovine. The average daily Cd intake ($\mu\text{g day}^{-1}$) was calculated as the ratio of the lifetime exposure (Eq. (2)) to the age of the animal expressed in days. This value is taken forward to the kinetic uptake model (Section 2.2.3).

The ingested amounts of feed and water were calculated with consumption patterns (Tables 2–4). The consumption pattern differed for calves born in summer or autumn (born in August–November: weaners reared outdoors during the following pasture season) versus calves born in winter or spring (born in December–June: weaners kept in the stable for about one year). Furthermore, different consumption patterns were used for beef and dairy cattle, and for

Table 2

Feed and water consumption patterns for calves and weaners.

Feed	Feed intake (kg DM/day)		
	Calves born in summer–autumn		Calves born in winter–spring
	Stable period	Pasture period	Stable period
Pasture grass	0	4.5	0
Silage grass	1.8	0	3
Silage maize	0.5	0	1
Concentrates	1.5	1.5	2
Water (l/day)	7	7	7

Table 3

Feed and water consumption patterns for dairy and beef yearlings.

Feed	Feed intake (kg DM/day)			
	Dairy cattle		Beef cattle	
	Stable period	Pasture period	Stable period	Pasture period
Pasture grass	0	9	0	8.5
Silage grass	4	0	3	0
Silage maize	4	0	3	0
Concentrates	0	0	3	1.5
Water (l/day)	25	25	25	25

the winter (stable period) and summer (pasture period) season. The length of the stable and pasture period was farm-dependent. The majority of the farmers allowed the cattle on the pastures from the beginning of April till the end of October (7 months pasture–5 months stable). The amounts of feed consumed were derived from Belgian literature to represent as much as possible the feeding strategies of Belgian farmers (ABKL, 2005; LV, 2008a, 2008b). The majority of the dairy cattle farmers in the current study used a maize/grass ratio of 63/37 (on a dry matter basis), while the majority of the beef cattle farmers used a ratio of 50/50. These ratios were used as “standard ratios”. Nevertheless, other ratios were applied as well (e.g. 0/100, 100/0, 80/20, ...). Whenever the farmers provided other ratios, this information was preferentially used over the standard ratios. Soil might be ingested in a passive process through the consumption of roughage with adhering soil dust, or in an active process during the pasture period (Fries et al., 1982). Since Cd concentrations were measured in pasture and silage grass and in maize, it was assumed that Cd in adhering soil was included in those measurements, and passive soil intake was not additionally accounted for. The amount of active soil ingestion during the pasture period was set equal to 2.4% of the dry matter grass intake (Fries et al., 1982).

Cadmium concentrations were measured in the different matrices (feed, water, and soil) for the 57 cases that were studied in depth. If animals received tap water (which was not sampled and analyzed), the concentration of Cd was set equal to the lowest measured Cd concentration in pump water ($0.01 \mu\text{g l}^{-1}$). It was assumed that during the pasture period, 50% of the water was consumed in the pasture and 50% was consumed in the stable. Cadmium concentrations in concentrates were set equal to $50 \mu\text{g kg}^{-1}$ (Römken et al., 2007).

2.2.3. Kinetic uptake model

The Nordberg–Kjellström model (Kjellström and Nordberg, 1978), which is a kinetic model of Cd metabolism in man, was used as the basis for a kinetic uptake model in bovines, as it has been obtained partly from animal experiments and because the metabolism of Cd is similar for several mammals (Neathery and Miller, 1975). A flow scheme is presented in Fig. 2. The model is simplified compared to the original Nordberg–Kjellström model in two ways: (1) Cd exposure through inhalation has been ignored because the animals have not

Table 4

Feed and water consumption patterns for dairy and beef cattle older than 2 years.

Feed	Feed intake (kg DM/day)			
	Dairy cattle		Beef cattle	
	Stable period	Pasture period	Stable period	Pasture period ^a
Pasture grass	0	9	0	13
Silage grass	4.4	3	5.5	0
Silage maize	7.5	1.3	5.5	0
Concentrates	5	2.5	0	2.5
Water (l/day)	80	80	45	45

^a Whether there is a pasture period for beef cattle depends on the age the animals reached (more than 3 years) and the management practice of the farmer.

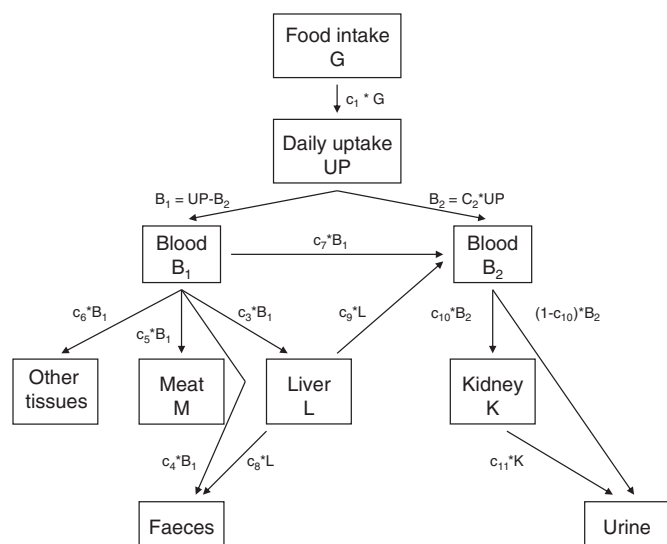


Fig. 2. Flow scheme of the kinetic uptake model for cadmium. Parameter values c_1 – c_{11} are explained in Section 2.2.3.

been exposed to increased Cd concentrations in air, and (2) the number of blood compartments has been reduced to two because it was not our intention to calculate the blood amount of Cd. On the other hand, meat has been added as an additional accumulation compartment. The model is based on the flow of amounts (μg) of Cd between the compartments.

Initial ranges of parameter values for the model (c_1 – c_{11} ; Table 5) were based on ruminant literature data and are clarified further below. Gastrointestinal absorption of Cd (c_1) is low in ruminants: Crout et al. (2004) estimated the absorption to be 0.118% and according to Van Bruwaene et al. (1982) it is 0.2% of the dose or less. About 14 days after dosing lactating Jersey cows, Neathery et al. (1974) retrieved 0.75% of the single ^{109}Cd dose in the total body, including the gastrointestinal tract and its contents. Based on these data, c_1 was initially assumed to be in the range 0.1–0.75%. The total amount of absorbed Cd (UP) goes to two blood compartments. The compartment B_1 represents blood plasma which is cleared from Cd within one day. The compartment B_2 represents circulating metallothionein. The fraction of absorbed Cd that binds directly on the circulating metallothionein (c_2) contributes to the Cd accumulation in kidneys (K) in the short-term. At longer term, Cd also redistributes from the liver (L) to the kidneys. Van Bruwaene et al. (1982) recovered 24% of a single oral ^{109}Cd administration in the kidneys after 131 days, compared to 50% in the liver. Therefore c_2 was initially assumed to be in the range 10–35% and c_3 , the distribution from the blood plasma to the liver, in the range 40–80%. Similar to Kjellström and Nordberg (1978) it was assumed that a proportion of Cd in the

blood plasma (c_4 : 5–50%) was excreted via the intestinal walls to the feces. The distribution of Cd from blood plasma to meat (M; c_5) and other tissues (e.g. bone, fat, skin, and milk; c_6) was initially based on data of Van Bruwaene et al. (1982; 2.1% in muscle and 16% in carcass excluding muscle) and Crout et al. (2004; 1% of GI absorbed dose in muscle and 3.5% of GI absorbed dose in bone, fat and skin). The fraction of Cd that goes from blood plasma to circulating metallothionein (c_7) was estimated at 0.4–1.5%. As ^{109}Cd was undetectable in plasma samples within the first 72 h after an oral administration (Crout et al., 2004), the sum of c_3 , c_4 , c_5 , c_6 and c_7 was set to be within the range 0.95–1. The major source of renal Cd in chronic exposure is derived from hepatic Cd, which is transported in the form of Cd-metallothionein (Chan and Cherian, 1993). This is a relatively slow process, and therefore c_9 was initially estimated to range from 0.0001 to 0.003 d^{-1} . Together with the excretion of Cd from the liver via bile to the feces (c_8 : 0–0.001 d^{-1}) this results in a half-time of Cd in liver between 0.5 and 19 years. During filtration of the Cd-metallothionein (present in B_2) through kidney glomeruli, a large fraction is reabsorbed in the renal tubules of animals (c_{10}) while the remaining is excreted directly in urine ($1 - c_{10}$). Part of the Cd that accumulates in the kidney is finally excreted in urine (c_{11}), but the half-time in kidney is longer than that in liver. Therefore c_{11} was set initially between 0.0001 and 0.0005 d^{-1} , corresponding to a half-time of 4 to 19 years. Finally, it was assumed that there was no redistribution from the different tissues towards the blood compartment B_1 .

The kinetic uptake model is described by a set of simultaneous first-order differential equations:

$$\text{UP}(t) = c_1 \cdot G(t) \quad (3)$$

$$B_1(t) = \text{UP}(t) - B_2(t) \quad (4)$$

$$B_2(t) = c_2 \cdot \text{UP}(t) \quad (5)$$

$$dB_1(t)/dt = \text{UP}_1(t) - (c_3 + c_4 + c_5 + c_6 + c_7)B_1(t) \quad (6)$$

$$B_2(t) = \text{UP}_2(t) + c_7 \cdot B_1(t) + c_9 \cdot L(t) \quad (7)$$

$$dM(t)/dt = c_5 \cdot B_1(t) \quad (8)$$

$$dL(t)/dt = c_3 \cdot B_1(t) - (c_8 + c_9) \cdot L(t) \quad (9)$$

$$dK(t)/dt = c_{10} \cdot B_2(t) - c_{11} \cdot K(t) \quad (10)$$

Live-weight of the animals (BW) was calculated as

$$\text{BW}(t) = 192.64 \cdot \ln(0.0175 \cdot t) \quad (11)$$

which was derived by fitting an exponential equation to data of the desired growth development during the breeding of bovines (ABKL, 2005). As this model results in live-weights below the birth weight of calves (~ 40 kg) for $t < 70$ days, it was corrected as

$$\text{BW}(t) = 40 + t/3.5 \quad (12)$$

as long as Eq. (11) resulted in live-weights below 40 kg.

Liver weight (LW), kidney weight (KW) and muscle weight (MW) were calculated as

$$\text{LW}(t) = 0.01 \cdot \text{BW}(t) \text{ with } \text{LW}(t)_{\max} = 8 \text{ kg} \quad (13)$$

$$\text{KW}(t) = 0.0025 \cdot \text{BW}(t) \text{ with } \text{KW}(t)_{\max} = 3 \text{ kg} \quad (14)$$

$$\text{MW}(t) = 0.35 \cdot \text{BW}(t) \quad (15)$$

The equations were programmed and iteratively solved in Microsoft Visual Basic 6.5 (Microsoft Corp. 2006). The time unit was one day and the input into the kinetic model was the average daily Cd

Table 5

Initial ranges of parameter values for Cd in the dynamic uptake model, and final values used for validation.

Coefficient	Definition	Initial range	Final value
c_1	GI absorption	0.1–0.75 %	0.3
c_2	Direct binding to metallothionein	10–35 %	32
c_3	Plasma to liver	40–80 %	75
c_4	Excretion via intestinal walls	5–50 %	7.2
c_5	Plasma to meat	1–10 %	7.3
c_6	Plasma to other tissues	5–25 %	10
c_7	Plasma to metallothionein	0.4–1.5 %	0.5
c_8	Liver to feces	0–0.001 d^{-1}	0.0005
c_9	Liver to metallothionein	0.0001–0.003 d^{-1}	0.0008
c_{10}	Metallothionein to kidney	80–98 %	85
c_{11}	Urinary excretion	0.0001–0.0005 d^{-1}	0.0002

intake (G; expressed in μg) calculated in the exposure model. Distribution towards other tissues, feces and urine were not calculated as such. The model was calibrated by comparing the calculated Cd concentrations in kidney, liver and meat with the measured concentrations in these tissues for the 57 animals for which detailed exposure calculations could be performed based on measured Cd concentrations in feed, drinking water and soil. To determine the best reasonable fit, the error sum of squares between log-transformed observed and calculated values was minimized for the three compartments (kidney, liver and meat).

2.2.4. Chain model validation

To validate the complete dynamic chain model (exposure model + kinetic uptake model), the 93 remaining animals were used. The available information on these animals was limited to the age of the animals (dates of birth and slaughtering), sex, type of the animal (dairy or beef cattle) and the locations where they had resided. The input values into the chain model were Cd concentrations in soil and drinking water, soil pH and soil organic matter content, which were all taken as typical regional values (Table 6) and Cd concentrations in concentrates ($50 \mu\text{g kg}^{-1}$). The regional values were selected based on data from Waegeneers et al. (1999), De Temmerman et al. (2003) and data from the current study. For animals originating from one of the three contaminated areas, input values were based on data of nearby farms that had been visited. From these data and the standard feed and water consumption patterns (Tables 2–4) the average daily Cd intake was calculated and taken forward to the kinetic uptake part of the model to calculate Cd concentrations in kidney, liver and meat. These calculated values were compared to the corresponding measured concentrations in the tissues of these animals.

3. Results and discussion

3.1. Analytical results

Table 7 shows the results of some chemical soil characteristics of pasture and arable soils. The mean, median and range of Cd concentrations were similar in both types of soils. The soils were sampled both in reference areas and in contaminated areas, which explains the large range in Cd concentrations. The soil Cd concentrations ranged from 0.12 to 2.0 mg kg^{-1} (average 0.5 mg kg^{-1}) in the reference areas and from 0.12 to 11 mg kg^{-1} (average 1.0 mg kg^{-1}) in the contaminated areas. The largest Cd concentrations (7 – 11 mg kg^{-1}) were found in soils from the Scheldt polder. The mean pH was slightly higher in arable soils than in pasture soils, which was significant in the contaminated areas but not in the reference areas (details not shown). The organic matter content was significantly lower in arable soils compared to pasture soils in both areas.

Table 6

Chain model input values per region: Cd concentrations in soils (mg kg^{-1}) and in drinking water ($\mu\text{g l}^{-1}$), pH and organic matter content (%) in pasture and arable soils.

Region		Soil Cd	pH		OM		Drinking water	
			Pasture	Arable	Pasture	Arable	Field	Stable
Zandstreek	East	0.34	5.5	5.7	4.6	2.8	0.15	0.01
	West	0.28	5.5	5.7	4.6	2.8	0.15	0.01
Zandleemstreek	East	0.3	5.7	5.7	5.4	1.7	0.15	0.01
	West	0.5	5.7	5.7	5.4	1.7	0.15	0.01
Kempen	Vochtig Haspengouw	0.3	5.7	5.7	5.4	1.7	0.15	0.01
	North	0.28	5.5	5.6	5.5	3.3	0.3	0.1
Leemstreek	East	0.5–1.0 ^a	5.5	5.6	5.5	3.3	0.45	0.18
	General	0.6	6.0	5.5	5.4	5.5	0.18	0.09
Weidestreek	Droog Haspengouw	1.0	6.0	5.5	5.4	5.5	0.18	0.09
	North	1.0	5.9	6.1	7.5	4.5	0.15	0.08
Condroz	South	0.36	5.9	6.1	7.5	4.5	0.15	0.08
		0.36	6.7	6.9	4.6	2.7	0.15	0.01
Les Ardennes		0.4	5.9	6.0	7.0	4.2	0.15	0.01

^a Kempen–east, but close to contaminated sites.

Table 7

Some chemical soil characteristics for pasture soils ($n = 127$) and arable soils ($n = 125$) on an air-dry weight basis.

	Pasture soils			Arable soils		
	Mean	Median	Range	Mean	Median	Range
[Cd] mg kg^{-1}	0.82	0.44	0.12–10.8	0.82	0.40	0.12–10.1
pH(H_2O)	5.9	5.8	4.7–7.5	6.1	6.1	4.9–8.1
OM ^a %	5.2	5.0	1.5–13.3	3.1	2.8	1.5–9.8

^a OM is the organic matter content.

Cadmium concentrations in pasture grass were significantly lower in spring compared to summer and autumn (Table 8) a trend that also has been reported by Loganathan et al. (1996). There were no significant differences in Cd concentrations between the three types of fodder (details not shown). The maximum level of Cd in animal feed of vegetable origin in the European Union is 1 mg kg^{-1} at a feed moisture content of 12%, which corresponds to 1.14 mg kg^{-1} on a dry weight basis (EU, 2002). This value was exceeded in 2% of the grass samples and in 6% of the maize silage samples.

Cadmium concentrations measured in well water and surface water ranged from 0.01 to $4.11 \mu\text{g l}^{-1}$. The mean was $0.44 \mu\text{g l}^{-1}$, the median $0.12 \mu\text{g l}^{-1}$. Cadmium concentrations in kidney, liver and meat have been reported in Waegeneers et al. (2009a). In the 57 cases that were studied in depth, Cd concentrations in kidney, liver and meat ranged respectively from 193 to $15,300 \mu\text{g kg}^{-1}$, from 49 to $2650 \mu\text{g kg}^{-1}$ and from 1 to $12 \mu\text{g kg}^{-1}$. In the remaining 93 animals, Cd ranged from 93 to $5870 \mu\text{g kg}^{-1}$, 29 to $888 \mu\text{g kg}^{-1}$ and 1 to $12 \mu\text{g kg}^{-1}$ in kidney, liver and meat respectively.

3.2. Modeling concepts

3.2.1. Bovine intake of trace elements

For each of the 57 animals that were studied in depth, lifetime Cd intake and mean daily Cd intake were calculated based on the locally measured Cd concentrations in feed, soil and water and the consumption patterns (Tables 2–4). The calculated lifetime Cd intake varied from 264 mg to 12 g . The mean daily Cd intake ranged from 0.5 mg day^{-1} to 13.6 mg day^{-1} . Fig. 3 shows the relative contribution of the different sources of Cd to the mean daily Cd intake for three scenarios: (1) a case with a low mean daily Cd intake, 0.7 mg day^{-1} (due to low levels of Cd in soil and feed), (2) a case with a high mean daily Cd intake, 3.1 mg day^{-1} (due to increased levels of Cd in soil and feed), and (3) a theoretical case based on median soil characteristics (cfr. Table 7) resulting in an intermediate daily Cd intake (1.2 mg day^{-1}). All three cases show that Cd in grass is the main source of daily Cd intake (60–75%). Whether pasture grass or grass silage is more important depends on the length of the stable period

Table 8

Summary statistics of Cd concentrations measured in locally grown fodder crops. The Cd concentrations are expressed on an oven dry weight basis.

		n	Mean mg kg ⁻¹	Median	Range
Grass	Spring	123	0.144	0.090	0.024–1.63
	Summer	118	0.217	0.141	0.029–1.39
	Autumn	113	0.228	0.118	0.034–2.22
	Silage/hay	53	0.190	0.122	0.033–0.856
Maize	Silage	84	0.287	0.107	0.006–3.70

and the type of cattle. Cadmium in maize accounts for 20 to 30% of the mean daily Cd intake. Active soil ingestion accounts for less than 5% and water intake accounts for less than 1% of the mean daily Cd intake.

3.2.2. Kinetic uptake model

Cadmium levels in kidneys, predicted for the 57 cases with the kinetic uptake model, ranged from 245 to 8580 $\mu\text{g kg}^{-1}$ (median: 1370 $\mu\text{g kg}^{-1}$) while the observed values ranged from 193 to 15,300 $\mu\text{g kg}^{-1}$ (median: 1560 $\mu\text{g kg}^{-1}$). Predicted Cd levels in liver ranged from 72 to 2220 $\mu\text{g kg}^{-1}$ (median: 229 $\mu\text{g kg}^{-1}$) while the observed values ranged from 49 to 2650 $\mu\text{g kg}^{-1}$ (median: 257 $\mu\text{g kg}^{-1}$). The range in predicted Cd levels in meat was 1–10 $\mu\text{g kg}^{-1}$, while the observed range was 1–13 $\mu\text{g kg}^{-1}$ (median values 2 and 1 $\mu\text{g kg}^{-1}$ respectively). The kinetic uptake model for Cd accounted for 47% of the variation in the observed kidney Cd concentrations, 43% of the variation in liver Cd concentrations and 21% of the variation in meat Cd concentrations when the parameter values as presented in Table 5, were used (Fig. 4). The slopes of the regression equations between log-transformed predicted and observed values were 0.87, 0.83 and 0.57 for kidney, liver and meat respectively. The intercepts of these equations did not differ significantly from zero for the kidney and liver compartments. The standard errors of the estimates were 0.31, 0.26 and 0.29 for kidney, liver and meat respectively, with the standard deviations of the observations being 0.42, 0.35 and 0.32 respectively. The regression between observed and predicted data in the liver compartment is influenced by two elevated liver Cd concentrations (2320 and 2650 $\mu\text{g kg}^{-1}$). These concentrations were rather exceptional as only three liver Cd concentrations in the whole dataset of 150 animals exceeded 1000 $\mu\text{g kg}^{-1}$. Therefore the performance of the kinetic

uptake model was also evaluated by excluding these two data points. As a result, the range in predicted liver Cd concentrations still reflected the observed range (predicted: 72–676 $\mu\text{g kg}^{-1}$; observed: 49–773 $\mu\text{g kg}^{-1}$) although the model accounted for only 24% of the variation in observed liver Cd concentrations. In general, the kinetic uptake model performed well: there is still unexplained variation in the individual data points, but the ranges and median Cd concentrations were well estimated. The model performed least for meat.

The estimated parameter values for excretion of Cd from liver and kidney result in half-time values of 1.5 and 9.5 years respectively. There is a large discrepancy with the half-times that can be calculated from data of Crout et al. (2004) for dairy cattle, i.e. 21 years for liver and more than 10⁶ years for kidney. It must be stressed that the latter values were accompanied by large uncertainties, as the half-time in kidney could as well be calculated to be 21.5 years. The longer the half-time, the smaller the probability that Cd levels in tissues decrease again when the diet is switched from contaminated to uncontaminated.

3.2.3. Chain model validation

The soil–plant transfer models were validated with independent data sets of paired measurements of soil Cd concentrations, soil characteristics and plant Cd concentrations (Smolders et al., 2007). The estimated Cd levels in grass ranged from 0.05 to 0.84 mg kg^{-1} (median 0.22 mg kg^{-1}) while the measured values ranged from 0.06 to 0.89 mg kg^{-1} (median 0.15 mg kg^{-1}). The estimated Cd levels in maize ranged from 0.02 to 2.8 mg kg^{-1} (median 0.24 mg kg^{-1}) while the measured values ranged from 0.04 to 6.8 mg kg^{-1} (median 0.31 mg kg^{-1}). These estimated Cd concentrations show acceptable agreement with the measured values.

Additional data were gathered from literature to validate specifically the kinetic part of the chain model. Johnson et al. (1981) fed six Hereford steers (336 kg live-weight) a diet containing Cd contaminated sewage sludge. After 106 days on the sludge diet the steers (~460 kg live-weight) were slaughtered and samples of muscle, liver and kidney were analyzed for Cd. Tissues were also collected at that time from six control steers fed the same diet without sludge. The average daily Cd intake was 1.20 mg d^{-1} for the control group and 94.9 mg d^{-1} for the sludge-fed group. The kinetic model described above was run on these data. The weight gain during the 106 days treatment period (~1.2 kg day^{-1}) was larger than the weight gain in the model calculations. The modeling results were therefore adjusted

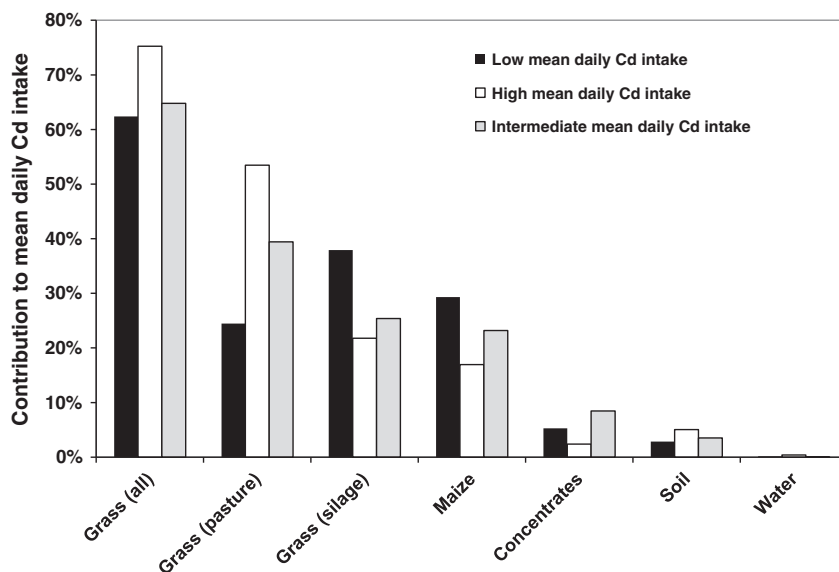


Fig. 3. Relative contribution of different sources of Cd to the mean daily Cd intake for two real-life cases (low and high mean daily Cd intake, 0.7 and 3.1 mg Cd day^{-1} respectively) and a theoretical case (intermediate daily Cd intake, 1.2 mg day^{-1}).

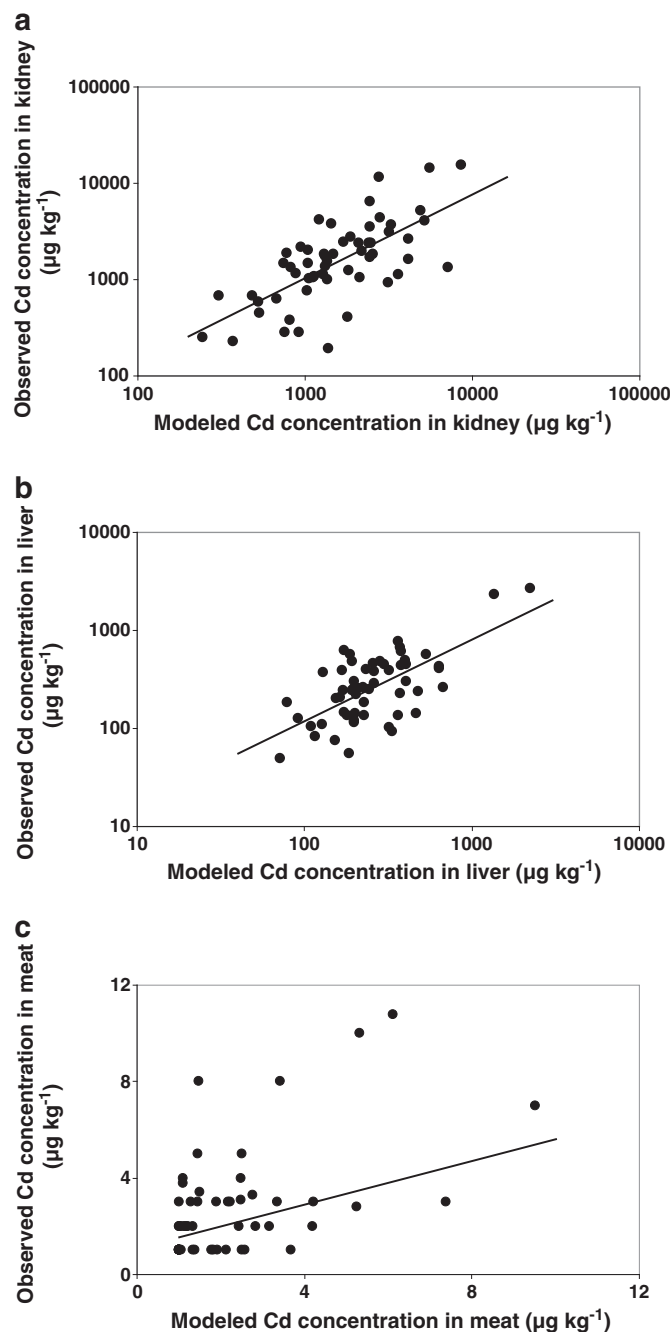


Fig. 4. Comparison of observed Cd concentrations in (a) kidney, (b) liver and (c) meat of the 53 bovines, used to derive the kinetic model parameters, with model predictions. The regression lines between observed and modeled data are indicated as straight lines.

for the 15% difference in weight at slaughter. The tissue Cd concentrations reported by Johnson and coworkers are expressed on a dry weight basis. To convert them to fresh weight data, moisture contents of 79.1% (kidney), 70.3% (liver) and 72.3% (meat) were used (Olsson et al., 2001). The model calculations overpredict the observed values in kidney and liver by a factor two to three, but meat concentrations seem to be accurately predicted (Table 9). Lamphere et al. (1984) fed 6–9 months old calves (184 kg live-weight) a diet supplemented with CdCl_2 for 60 days after which the calves were slaughtered and samples of kidney cortex, liver and muscle were collected. The average daily Cd intake varied with live-weight as the calves were fed a diet containing $\sim 50 \text{ mg Cd kg}^{-1}$ feed at a rate of 2% of body weight per day. The observed Cd concentrations were compared with predictions from the kinetic model. Hereto kidney

Table 9

Comparison of dynamic model predictions for Cd concentrations in kidney, liver and meat with experimental data.

Reference	Tissue	Cd concentration ($\text{mg kg}^{-1} \text{fw}$)	
		Model prediction	Observed value ^a
Johnson et al. (1981)	Kidney—control group	0.45	0.25 ± 0.03
	Kidney—sludge-fed group	7.9	3.04 ± 0.26
	Liver—control group	0.14	0.06 ± 0.003
	Liver—sludge-fed group	3.6	1.46 ± 0.13
	Meat—control group	≤ 0.001	< 0.003
Lamphere et al. (1984)	Meat—sludge-fed group	0.01	0.01 ± 0.003
	Kidney—baseline group	0.19	0.22 ± 0.08
	Kidney—treated group	18.3	26.6 ± 3.3
	Liver—baseline group	0.08	0.06 ± 0.01
	Liver—treated group	8.76	8.77 ± 0.42
Smith et al. (1991)	Meat—baseline group	≤ 0.001	0.004 ± 0.001
	Meat—treated group	0.025	0.016 ± 0.003
	Kidney—low Cd	4.94	3.92
	Kidney—high Cd	24.19	27.62
	Liver—low Cd	1.49	0.60
	Liver—high Cd	7.36	4.28

^a Mean \pm standard error.

cortex concentrations were recalculated to whole kidney concentrations assuming that the Cd concentration in the cortex is 37% higher than the renal average (Olsson and Oskarsson, 2001). Given the uncertainties on the observed data, the model performed very well (Table 9). In a study of Smith et al. (1991), Holstein heifers averaging 13 months of age (340 kg live-weight) were fed a low-Cd diet ($0.025 \text{ mg Cd kg}^{-1}$ body weight) or a high-Cd diet ($0.125 \text{ mg Cd kg}^{-1}$ body weight) during on average 554 days. Cadmium concentrations in kidney, liver and muscle, which were again expressed on a dry weight basis, were converted to fresh weight concentrations with the moisture contents of Olsson et al. (2001). The Cd concentrations in muscle were not considered reliable as even in cows that were not fed additional Cd, the muscle concentrations were $0.02\text{--}0.05 \text{ mg Cd kg}^{-1}$ fresh weight, which is high for unpolluted muscle tissues (López-Alonso et al., 2000 and references therein; Waegeneers et al., 2009a). The modeling results were in close agreement to the observed Cd concentrations in kidney, but the liver concentrations were over-predicted by a factor 1.7 to 2.5 (Table 9). Given all these data, the kinetic part of the dynamic chain model seems to perform reasonably well over a wide range of dietary Cd intake levels.

The complete dynamic chain model was validated with the 93 remaining animals. The modeled average daily Cd intake by these animals varied from $0.249 \text{ mg day}^{-1}$ to 4.26 mg day^{-1} . The average daily Cd intake values for each of the 93 animals were taken forward to the kinetic uptake model to predict the corresponding Cd concentrations in kidney, liver and meat. The results of these modeling calculations are presented in Fig. 5. In general, the chain model performed well with 90% of the data being predicted within a factor 3 of the observed data. Predicted Cd levels in kidneys ranged from 97 to 6600 µg kg^{-1} (median: 1520 µg kg^{-1}) while the observed values ranged from 93 to 5870 µg kg^{-1} (median: 954 µg kg^{-1}). Predicted Cd levels in liver ranged from 34 to 740 µg kg^{-1} (median: 208 µg kg^{-1}) while the observed values ranged from 29 to 888 µg kg^{-1} (median: 167 µg kg^{-1}). The range in predicted Cd levels in meat was $1\text{--}6.7 \text{ µg kg}^{-1}$, while the observed range was $1\text{--}12 \text{ µg kg}^{-1}$ (median values 1.6 and 1 µg kg^{-1} respectively). The chain model accounted for 65% of the variation in the observed kidney Cd concentrations, 42% of the variation in liver Cd concentrations and 32% of the variation in meat Cd concentrations. Part of the remaining uncertainty might be attributed to the fact that many animals were not reared at a single location but at two to three farms spread over the country. Soil characteristics, and hence Cd concentrations in feed, vary between regions. Moving animals from one region to another might significantly influence the lifetime and average daily Cd intake.

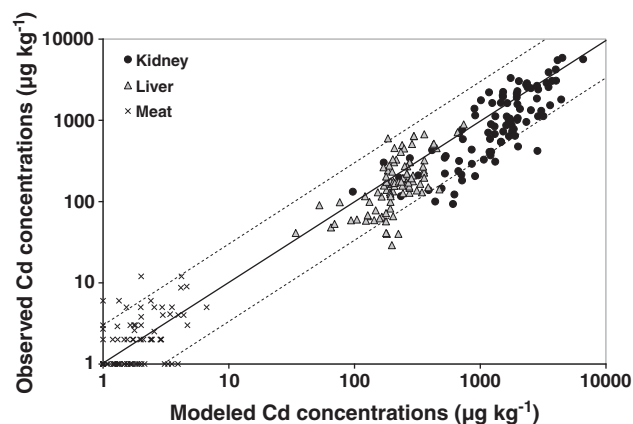


Fig. 5. Comparison of observed and modeled tissue Cd concentrations for the 97 bovines used to validate the chain model. The 1:1 line is indicated as a straight line, the dotted lines represent observed/modeled = 3 and 0.33.

4. Discussion

In contrast to the dynamic modeling approach used in the present study, cadmium concentrations in animal tissues can also be predicted by using equilibrium transfer coefficients, defined as the equilibrium ratio of the fresh weight Cd concentration in tissue to the daily Cd intake (F , d kg^{-1}), or concentration ratios, defined as the equilibrium ratio between the Cd concentration in the tissue and its concentration in the feed ingested (CR , unitless) (Howard et al., 2009a). The advantage of transfer coefficients and concentration ratios is that they are easy to apply, but the disadvantage is that the range of these values can be large. Recommended equilibrium transfer coefficients for Cd to cow meat range from $1.5 \cdot 10^{-4}$ to $6.0 \cdot 10^{-2} \text{ d kg}^{-1}$ (Howard et al., 2009b) and concentration ratios range from $2.3 \cdot 10^{-3}$ to $3.5 \cdot 10^{-1}$ (Howard et al., 2009a). These ranges of transfer coefficients and concentration ratios have been applied to the 93 animals used for model validation. As only F and CR values for meat are available, a weighted mean cadmium concentration ratio kidney/liver/meat of 134/31/1 was used to calculate Cd concentrations in kidney and liver (Franz et al., 2008). The lowest F and CR values largely underpredict the observed data, while the largest F and CR data largely overpredict the observed data (Table 10). Furthermore the span between lowest and highest kidney Cd concentration was a factor 63 in the observed data, but only a factor 17 when using transfer coefficients and a factor 6 when using concentration ratios. Concentration ratios do not take into account variations in dry matter intake between animals, and neither concentration ratios nor transfer coefficients take into account the age of the animals. Franz et al. (2008) integrated the age of animals in the calculation of Cd concentrations in kidney and liver by multiplying a linear biotransfer rate (BTR ; kg^{-1}) with the daily Cd intake and animal age. The model of Franz and coworkers represents a considerable improvement in predicting the Cd contamination in cattle tissues because cattle consumption patterns are used to calculate the daily Cd intake and because the age-dependent increase in tissue Cd levels is considered. The model of Franz was also applied on the 93 animals used for model validation. The predicted Cd intake (range 0.18 – 4.63 mg day^{-1}) was similar to the Cd intake predicted in the current study (0.25 – 4.26 mg day^{-1}), as should be since similar consumption patterns were used. Nevertheless, final Cd concentrations in the different tissues were underestimated by the model (Table 10). This should be due to the choice of the BTR . Crout et al. (2004) described the transfer of Cd to dairy cattle with a kinetic model. The parameters for the model were derived from an experiment where a single oral administration of ^{109}Cd was given to sheep and adjusted according to the ratio of the metabolic live-weights of sheep to that of cattle. The resultant model was validated through an experiment in which three

Table 10

Performance of different approaches (transfer coefficients F ; concentration ratios CR ; chain model by Franz et al. (2008); current chain model) to estimate cadmium levels in bovine kidney, liver and meat.

	Kidney [Cd] ($\mu\text{g kg}^{-1}_{\text{fw}}$)		Liver [Cd] ($\mu\text{g kg}^{-1}_{\text{fw}}$)		Meat [Cd] ($\mu\text{g kg}^{-1}_{\text{fw}}$)	
	Median	Range	Median	Range	Median	Range
Observed data	954	93–5870	167	29–888	1.0	1–12
$F = 1.5 \cdot 10^{-4}$	24	5–86	6	1–20	0.2	0.04–0.6
$F = 6.0 \cdot 10^{-2}$	9790	2000–34,300	2270	462–7920	73	15–256
$CR = 2.3 \cdot 10^{-3}$	42	19–120	10	4–28	0.3	0.1–0.9
$CR = 3.5 \cdot 10^{-1}$	6330	2920–18,300	1460	674–4230	47	22–136
Franz et al. (2008)	164	6–993	31	1–188	1.2	0.05–7.4
Current model	1520	97–6600	208	34–740	1.6	1.0–6.7

dairy cows were given a single intraruminal administration of ^{109}Cd and measurements were made of the subsequent ^{109}Cd concentrations in different body tissues (and milk) 28 days after dose application. The model was an improvement in the ability to predict the Cd contamination in cattle tissues from Cd intake rates. Unfortunately, the uncertainty on predicted tissue Cd concentrations was very large for several tissues, the standard error equaled up to 180 times the mean predicted concentration. Furthermore Crout et al. (2004) indicated themselves that their model might not accurately predict transfer at higher Cd intake rates. This limitation has been overcome in the current kinetic model as it has been calibrated and validated with a range of Cd intake rates from 0.25 to 14 mg d^{-1} . The model of Crout et al. (2004) was too complex to be applied on the 93 animals used for model validation. From the different approaches that have been applied to the 93 animals to predict tissue Cd concentrations, the current dynamic chain model performed best with the median and ranges corresponding well with observed data (Table 10). Also the span between lowest and highest predicted kidney Cd concentration (a factor 68) corresponded well with the span in the observed data (a factor 63). The current chain model combines the advantages of the model of Franz et al. (2008) with the ideas of kinetic modeling. Although the model of Franz et al. (2008) is very valuable, the performance of the current chain model was better than calculations that rely on the linear transfer of Cd to animal tissues when applied on the 93 animals used for validation (Table 10).

The advantage of the current dynamic chain model, which consists of a series of coupled models in the soil–feed–animal tissue chain, is that depending on the available input data (soil data, feed data or daily intake data), the model calculations can start at different levels. Further the chain model is valid for both dairy and beef cattle and for animals between 0.5 and 11.5 years of age.

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References

- ABKL. Succesvolle opfok van jongvee op het melkbedrijf. Brussel: Administratie Beheer en Kwaliteit Landbouwproductie (ABKL). Ministerie van de Vlaamse Gemeenschap; 2005 (in Dutch).
- Andersen A, Hovgård Hansen H. Cadmium and zinc in kidneys from Danish cattle. Nord Vet Med 1982;34:340–9.
- Beresford NA, Mayes RW, Crout NMJ, Maceachern PJ, Dodd BA, Barnett CL, et al. Transfer of cadmium and mercury to sheep tissues. Environ Sci Technol 1999;33:2395–402.
- Chan HM, Cherian MG. Mobilization of hepatic cadmium in pregnant rats. Toxicol Appl Pharmacol 1993;120:308–14.

- Crout NMJ, Beresford NA, Dawson JM, Soar J, Mayes RW. The transfer of ^{73}As , ^{109}Cd and ^{203}Hg to the milk and tissues of dairy cattle. *J Agric Sci* 2004;142:203–12.
- De Temmerman L, Vanongeval L, Boon W, Hoenig M, Geypens M. Heavy metal content of arable soils in northern Belgium. *Water Air Soil Pollut* 2003;148:61–76.
- EU. Directive 2002/32/EC of the European parliament and the council of 7 May 2002 on undesirable substances in animal feed. *Off J Eur Comm* 2002;L140/10.
- Franz E, Römkens P, van Raamsdonk L, van der Fels-Klerx I. A chain modeling approach to estimate the impact of soil cadmium pollution on human dietary exposure. *J Food Protect* 2008;71:2504–13.
- Fries GF, Marrow GS, Snow PA. Soil ingestion by swine as a route of contaminant exposure. *Environ Toxicol Chem* 1982;1:201–4.
- Howard BJ, Beresford NA, Barnett CL, Fesenko S. Quantifying the transfer of radionuclides to food products from domestic farm animals. *J Environ Radioact* 2009a;100:767–73.
- Howard BJ, Beresford NA, Barnett CL, Fesenko S. Radionuclide transfer to animal products: revised recommended transfer coefficient values. *J Environ Radioact* 2009b;100:263–73.
- Jackson ML. Soil chemical analysis. Englewood Cliffs, NJ: Prentice Hall; 1958.
- Johnson DE, Kienholz EW, Baxter JC, Spangler E, Ward GM. Heavy metal retention in tissues of cattle fed high cadmium sewage sludge. *J Anim Sci* 1981;52:108–14.
- Kjellström T, Nordberg G. A kinetic model of cadmium metabolism in the human being. *Environ Res* 1978;16:248–69.
- Lamphere DN, Dorn RC, Reddy CS, Meyer AW. Reduced cadmium body burden in cadmium-exposed calves fed supplemental zinc. *Environ Res* 1984;33:119–29.
- Lindén A, Olsson I-M, Oskarsson A. Cadmium levels in feed components and kidneys of growing/finishing pigs. *J AOAC Int* 1999;82:1288–97.
- Loganathan P, Hedley MJ, Gregg PEH, Currie LD. Effect of phosphate fertiliser type on the accumulation and plant availability of cadmium in grassland soils. *Nutr Cycl Agroecosys* 1996;47:169–78.
- Loganathan P, Louie K, Lee J, Hedley MJ, Roberts AHC, Longhurst RD. A model to predict kidney and liver cadmium concentrations in grazing animals. *N Z J Agric Res* 1999;42:423–32.
- López-Alonso M, Benedito JL, Miranda M, Castillo C, Hernández J, Shore RF. Toxic and trace elements in liver, kidney and meat from cattle slaughtered in Galicia (NW Spain). *Food Addit Contam* 2000;17:447–57.
- LV. Voeding van runderen van het Belgisch witblauwe ras. Brussel: Vlaamse Overheid, Departement Landbouw en Visserij (LV), Afdeling Duurzame Landbouwwontwikkeling, Voorlichting en Vorming; 2008a (in Dutch).
- LV. Rundvleesproductie: voeding van vleesstieren. Brussel: Vlaamse Overheid, Departement Landbouw en Visserij (LV), Afdeling Duurzame Landbouwwontwikkeling; 2008b (in Dutch).
- Neathery MW, Miller WJ. Metabolism and toxicity of cadmium, mercury, and lead in animals: a review. *J Dairy Sci* 1975;1767–81.
- Neathery MW, Miller WJ, Gentry RP, Stake PE, Blackmon DM. Cadmium-109 and methyl mercury-203 metabolism, tissue distribution, and secretion into milk of cows. *J Dairy Sci* 1974;57:1177–83.
- Olsson I-M, Oskarsson A. Sampling of kidneys from cattle and pigs for cadmium analysis. *Analyst* 2001;126:114–20.
- Olsson I-M, Jonsson S, Oskarsson A. Cadmium and zinc in kidney, liver, muscle and mammary tissue from dairy cows in conventional and organic farming. *J Environ Monit* 2001;3:531–8.
- Römkens PFAM, Zeilmaker MJ, Rietra RPJJ, Kan CA, van Eijkeren JCH, van Raamsdonk LWD, et al. Blootstelling en opname van cadmium door runderen in de Kempen: een modelstudie. Alterra-rapport 1438 Wageningen: Alterra; 2007 (in Dutch).
- Smith RM, Leach RM, Muller LD, Griel Jr LC, Baker DE. Effects of long-term dietary cadmium chloride on tissue, milk, and urine mineral concentrations of lactating cows. *J Anim Sci* 1991;69:4088–96.
- Smolders E, Janssen G, Van Laer L, Ruttens A, Van Gronsveld J, Römkens P, et al. Teeltadvies voor de landbouw in kader van het Interreg project BeNeKempen. Internal Report OVAMMechelen: OVAM; 2007 (in Dutch).
- Van Bruwaene R, Gerber GB, Kirchmann R, Colard J. Transfer and distribution of radioactive cadmium in dairy cows. *Int J Environ Stud* 1982;19:47–51.
- Waegeneers N, Smolders E, Merckx R. A statistical approach for estimating the radiocaesium interception potential of soils. *J Environ Qual* 1999;28:1005–11.
- Waegeneers N, Pizzolon J-C, Hoenig M, De Temmerman L. Accumulation of trace elements in cattle from rural and industrial areas in Belgium. *Food Addit Contam* 2009a;26:326–32.
- Waegeneers N, Pizzolon J-C, Hoenig M, De Temmerman L. The European maximum level for cadmium in bovine kidneys is in Belgium only realistic for cattle up to 2 years of age. *Food Addit Contam* 2009b;26:1239–48.